

## **REMARKS**

Reconsideration of this application is respectfully requested in view of the foregoing amendments and the following remarks.

### **Priority**

As the Examiner points out, this application claims priority to PCT/US00/19007 ('007 herein) filed on July 13, 2000 which claims priority to provisional application serial No. 60/143,711 filed on July 13, 1999. The Examiner requests clarification as to whether the present application is a PCT 371 National Stage application of the '007 PCT application or a continuation. On October 17, 2002, the PCT Legal Office notified Applicants that the present application would be treated as filed under 35 U.S.C. 111(a).

### **Sequence Listing**

The Examiner indicates that the history of sequence listing filings in this application is confusing. As the Examiner points out, the first sequence listing filed on May 6, 2003 had four sequences. These polypeptide sequences represent site directed mutagenesis of the *cbhl* gene resulting in the replacement of asparagines 45, 270, and/or 384 with alanine. While the sequence listing filed March 20, 2007 no longer includes those entire polynucleotide sequences, Table 4 shows polynucleotide sequences with SEQ ID NOs 71, 74, and 77 which represent the mutation sites in the *cbhl* gene, numbered by the amino acid location as designated from the start of the mature protein (*i.e.*, without the signal sequence). The *cbhl* gene can contain any combination of the three mutations.

On March 8, 2004, Applicants submitted a sequence listing with 120 sequences which was subsequently deemed defective by the Patent Office. On June 21, 2004, Applicants resubmitted the sequence listing, again with 120 sequences. On August 24, 2004, an Office Action indicated sequences shown in the claims were not represented by the sequence listing, and in response, Applicants submitted a sequence listing containing 96 sequences on November 26, 2004. In conjunction with the sequence listing, Applicants explained in their response that the sequence listing was submitted to correct a clerical error in which the open reading frame of SEQ ID NO: 4 was inadvertently offset in the previous submission. This required a corresponding revision of the SEQ ID NOs in Tables 2-5. On June 22, 2006, Applicants submitted a replacement specification which necessitated the filing of still another sequence

listing. This sequence listing was filed on March 30, 2007 and reflects the 97 sequences shown in the specification. The Table below provides a showing of support for each of the 97 sequences (submitted in the March 30, 2007 Sequence Listing) in the originally filed specification. As can be seen, none of these sequences had sequence identifiers in the original specification, though each sequence is fully supported by the original specification.

Table 1. Sequence Support found in Specification filed January 14, 2002

SEQ ID NO:	Support in Paragraphs and/or Figures of January 14, 2002 Specification
1	Coding sequence for the linker region shown in Figure 4
2	Linker region amino acid sequence shown in Figure 4
3	Linker region nucleic acid sequence shown on page 15, last paragraph
4	Coding sequence for the <i>Trichoderma reesei cbh1</i> gene shown in Figure 1
5	Amino acid sequence for <i>Trichoderma reesei</i> CBH1 protein, encoded by SEQ ID NO: 4, and described on page 1, third paragraph
6-8	CBH1 variants: CBHI-N45A, CBHI-N270A, and CBHI-N384A described on page 3, third paragraph
9 and 97	Primers for the <i>T. reesei cbh1</i> gene shown on page 4, last paragraph
10-51	Native nucleic acid sequences and mutagenic sense and anti-sense strands shown in Table 2, pages 16-17
52-54	Native nucleic acid sequences and mutagenic sense and anti-sense strands shown in Table 3, page 17
55-69	Native nucleic acid sequences and mutagenic sense and anti-sense strands shown in Table 3b, page 17
70-78	Native nucleic acid sequences and mutagenic sense and anti-sense strands shown in Table 4, page 18
79-96	Native nucleic acid sequences and mutagenic sense and anti-sense strands shown in Table 5, pages 19

The Examiner requests a paper copy of the electronic Sequence Listing submitted on March 20, 2007. Enclosed is a paper copy of that same Listing. The contents of the electronic copy and the paper copy are the same. 37 C.F.R. § 1.821(f).

#### **Amendments to the Claims**

All references below to support for claim amendments refer to Applicants' specification as submitted in Appendix A, filed June 22, 2006, unless stated otherwise. Claims 6, 7, 9-16, 20-22, and 24-30 are pending. Claims 15, 16, 27, and 28 were previously withdrawn from consideration. Claim 24 is cancelled herein. This response amends claims 6, 7, 9-13, 20-22, and 26. Specifically:

Claim 6 is amended to include the “functions” encompassed by the term “functionality.” This amendment is supported by Applicants’ specification at, for example, page 2, third paragraph, and Tables 2 and 5 (thermal tolerance); page 2, fourth paragraph (enzymatic activity/catalytic activity); page 3, third paragraph, and Table 4 (reduced glycosylation); page 15, first paragraph, and Table 3 (peptide strain); and Table 3b (product inhibition).

Claim 7 is amended to indicate the functionality improved by the proline substitution at position 8. This amendment is supported by Applicants’ specification at, for example, Table 2.

Claims 20-22 are amended to reflect the functionality language of claim 7 from which the claims depend. This amendment is supported by Applicants’ specification at page 15, third full paragraph.

Claim 26 is amended to point out that the proline is substituted in the place of the serine at position 8 of SEQ ID NO: 5. This amendment is supported by Applicants’ specification at, for example, Table 2 and the paragraph spanning pages 15 and 16.

Claims 6, 9, 10, 11, 12, 13, and 14 are amended to make the language of the claims more clear by making minor grammatical changes.

New claims 31-35 are directed to a variant cellobiohydrolase having improved thermostability relative to the wild-type cellobiohydrolase. This protein has a substitution of proline at position 8, and can comprise further mutations as provided in dependent claims 32-35. These claims are supported by Applicants’ specification at, for example, Tables 2-5 and page 15, third full paragraph.

Applicants submit that none of these claim amendments introduce new matter.

### **Response to Examiner’s Arguments**

#### **A. Claim Rejections under 35 U.S.C. § 112, Second Paragraph**

Claims 6 and 26 are rejected as being indefinite for failing to particularly point out and distinctly claim the subject matter of the invention. Specifically, the Examiner finds that it is difficult to determine from the language of the claim whether the mutated *cbh1* gene encodes a protein with an amino acid sequence identical to that of SEQ ID NO: 5 with the exception of a proline at position 8 or instead encodes a protein different at all positions except for proline at position 8. As explained below, only one interpretation is possible.

Figure 1 shows the coding sequence for the *cbh1* gene (SEQ ID NO: 4) and in lower case letters, identifies the signal sequence. An exemplary mutation (and the subject of the election) is the substitution of proline for serine at position 8 of the CBH1 protein (SEQ ID NO: 5). The original triplet encoding the serine can be seen in Figure 1 of Applicants' specification, counting from the end of the signal sequence, at the 8th triplet, TCG (see also Table 2). With respect to the serine at position 8 in SEQ ID NO: 5, substitution with proline at the same position improves cellobiohydrolase functionality. Thus, any interpretation of the claims that concludes the variant cellobiohydrolase shares only the proline at position eight with the wild-type cellobiohydrolase fails to take into account the actual wild-type cellobiohydrolase sequence (*i.e.*, that there is no proline at position eight of the mature wild-type protein). For at least these reasons, Applicants respectfully request withdrawal of this rejection.

Claims 6, 7, 9, 10, 20-22, 24-26, and 28-30 are rejected for failing to particularly point out and distinctly claim the subject matter of the invention. Specifically, the Examiner finds the phrase "for improving the functionality of the variant" unclear as to whether the functionality "refers to the hydrolytic action, temperature stability, pH stability, etc." Claim 6 is amended to include the functions improved by cellobiohydrolase mutations, specifically thermostability, enzymatic activity, catalytic activity, product inhibition, glycosylation, and/or peptide strain. Applicants believe this amendment makes the claim more clear and addresses the Examiner's concern.

Claims 20-22 are rejected for failing to particularly point out and distinctly claim the subject matter of the invention. Specifically, the Examiner states that the word "thermostability" has insufficient antecedent basis. Claims 6 and 7 are presently amended to provide proper antecedent basis, thus addressing the Examiner's concern.

B. Claim Rejections under 35 U.S.C. § 112, First Paragraph

Claims 6, 7, 9, 10, 20-22, 25-26, and 29-30 are rejected for lack of enablement. Specifically, the Examiner's interpretation of claim 6 to include "any polynucleotide encoding any amino acid sequence, having cellobiohydrolase activity but for a proline at position 8" results in an enormous number of potential sequences. As shown above, this interpretation cannot be correct as a wild-type protein represented by SEQ ID NO: 5 always has a serine at position 8. Claim 6 is directed to a nucleic acid molecule encoding a variant cellobiohydrolase mutated with respect to a wild-type cellobiohydrolase encoded by SEQ ID NO: 5. The mutation

improves the functionality of the variant cellobiohydrolase relative to the wild-type cellobiohydrolase in terms of thermostability, enzymatic activity, catalytic activity, product inhibition, glycosylation, and/or peptide strain. With the proper claim interpretation in mind, it is Applicants' belief that a variant polynucleotide encoding a polypeptide mutated relative to SEQ ID NO: 5, and particularly mutated with a proline substitution at position 8 of the mature protein, having improved cellobiohydrolase functionality (thermostability, enzymatic activity, catalytic activity, product inhibition, glycosylation, and/or peptide strain), is sufficiently enabled by Applicants' specification.

C: Allowability of Claims 11-14

Applicants appreciate the Examiner's finding claims 11-14 allowable.

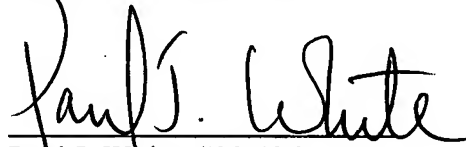
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CONCLUSION

For the reasons set forth above, Applicants respectfully submit that the claims are allowable and reconsideration and issuance of a notice of allowance are respectfully requested. If it would be helpful to obtain favorable consideration of this case, the Examiner is encouraged to call and discuss this case with the undersigned.

This constitutes a request for any needed extension of time and an authorization to charge all fees therefore to deposit account No. 14-0460 if not otherwise specifically requested. The undersigned hereby authorizes the charge of any required fees not included or any deficiency of fees submitted herewith to be charged to deposit account No. 14-0460.

Respectfully submitted,



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